

<b>7 EXTRACTION AND DERIVITIZATION</b>	Page 1 of 3
<div> <b>Division of Forensic Science</b>  <b>TOXICOLOGY TRAINING MANUAL</b> </div>	Amendment No.:
	Effective Date: 26-March-2004
<div> <p style="text-align: center;"><b>7 EXTRACTION AND DERIVITIZATION</b></p> <p><b>7.1 Objectives</b></p> <p>7.1.1 Understand the theoretical and practical aspects of extractions.</p> <p>7.1.2 Become familiar with various types of extractions.</p> <p>7.1.3 Perform different types of derivatization and comprehend why, when and how to use them.</p> <p>7.1.4 Extract representative compounds (basic, acidic &amp; neutral) from various matrices.</p> <p><b>7.2 Estimated Time:</b> Two months, part time</p> <p><b>7.3 Methods of Instruction</b></p> <p>7.3.1 Lecture</p> <p style="padding-left: 20px;">7.3.1.1 Principles of extraction</p> <p style="padding-left: 20px;">7.3.1.2 Henderson-Hasselbach equation, acid base equilibrium</p> <p style="padding-left: 20px;">7.3.1.3 Buffers and ionization</p> <p style="padding-left: 20px;">7.3.1.4 Extraction</p> <p style="padding-left: 20px;">7.3.1.5 Liquid-liquid extraction</p> <p style="padding-left: 20px;">7.3.1.6 Solid phase extraction (SPE)</p> <p style="padding-left: 20px;">7.3.1.7 Specimen preparation (dilution, internal standard, derivatization)</p> <p>7.3.2 Literature Review</p> <p style="padding-left: 20px;">7.3.2.1 Solid Phase Extraction Techniques (United Chemical Technologies).</p> <p style="padding-left: 20px;">7.3.2.2 Toxicology Technical Procedures Manual</p> <p style="padding-left: 20px;">7.3.2.3 Moffat, A.C., editor. <i>Clarke's Analysis of Drugs and Poisons</i>, 3<sup>rd</sup> edition. London: The Pharmaceutical Press, 2004 pp 80-108.</p> <p style="padding-left: 20px;">7.3.2.4 Stewart, C. P. &amp; Stolman, A. <i>Toxicology: Mechanisms and Analytical Methods</i>. Vol. 2, 1961, Academic Press.</p> <p style="padding-left: 20px;">7.3.2.5 <i>Handbook of Analytical Derivatization Reactions</i>, Daniel R. Knapp, John Wiley, New York, 1979.</p> <p style="padding-left: 20px;">7.3.2.6 Pierce Catalog (Pierce Endogen) 2001-2002, <i>GC Derivatization and Labware</i>, Pages 497 – 526.</p> <p>7.3.3 Demonstration</p> <p style="padding-left: 20px;">7.3.3.1 The following extraction and derivatization techniques will be observed from beginning to end and notes will be taken by the Trainee:</p> </div>	

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7.3.3.1.1	Liquid-liquid extraction	
7.3.3.1.2	Solid phase extraction	
7.3.3.1.3	Sample derivatization	
7.3.4	Laboratory exercises	
7.3.4.1	Perform an SPE extraction of cocaine/cocaethylene/benzoylecgonine calibrators. Derivatize and save extracts for analysis by GCMS in Section 8 (Gas Chromatography).	
7.3.4.2	Repeat extraction of cocaine/cocaethylene/benzoylecgonine calibrators using the liquid-liquid extraction procedure. Derivatize and save extracts for analysis by GCMS in Section 8 (Gas Chromatography).	
7.3.4.3	Extract and derivatize the acidic drugs present in the barbiturate standards using the liquid-liquid extraction procedure. Save extracts for analysis by GC/NPD in Section 8 (Gas Chromatography).	
<b>7.4</b>	<b>Evaluation</b>	
7.4.1	Written Exam	
7.4.1.1	This will be administered as a "take home" exam.	
7.4.2	Laboratory Competency Testing	
7.4.2.1	Solid phase extraction - a series of 10 previously analyzed blood specimens will be presented to the Trainee for cocaine quantitation. Results for controls and unknowns must agree within 20% of previous results per the Toxicology Technical Procedures Manual.	
7.4.2.2	Liquid-liquid extraction - a series of 10 previously analyzed blood specimens will be presented to the Trainee for a base screen. Save extracts for analysis in Section 8. Qualitative findings must agree with previous results.	
7.4.3	Courtroom Exercise	
7.4.3.1	The Trainee must be capable of answering questions on this Module such as would be expected in a courtroom scenario.	
<b>7.5</b>	<b>Examination Questions</b>	
7.5.1	Describe liquid-liquid and solid-phase extractions stating the advantages and disadvantages of each type.	
7.5.2	List and describe chemical forces which drive the movement of solute between aqueous and organic phases.	
7.5.3	Explain the effects of pH on extractions.	
7.5.4	List at least three different types of SPE sorbents, and how they interact with the substances being extracted.	
7.5.5	List and explain the typical steps in an SPE procedure.	
7.5.6	Define the following terms: matrix; functional group; polarity; solvents; pH; pKa; Henderson-Hasselbach equation; basic, acidic, neutral and amphoteric molecules; conjugate acid, conjugate base; internal standard; external standard.	
7.5.7	Describe silylation, methylation, and acylation.	

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7.5.8 Describe and/or draw the derivative formed using the Toxicology Technical Procedures Manual for morphine, benzoylecgonine, butalbital, amphetamine and tetrahydrocannabinol. ♦ End	